Bovine Babesiosis

Disease Monograph Series – 14

Parasite | Protozoa | *Babesia* | *B. bovis* | *B. bigemina* | Cattle

IDRC | Bartay

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Acronyms

ACIAR	Australian Center for International Agricultural Research
AI	Avian influenza
APMV	Avian paramyxovirus
AU	African Union
AU-IBAR	African Union InterAfrican Bureau For Animal Resources
AU PANVAC	AU Pan African Veterinary Vaccine Centre
BSL-3	Biosafety level 3 (Laboratory designation required for challenge studies)
DIVA	Differentiating infected from vaccinated animals (strategy)
ELISA	Enzyme-linked immunosorbent assay
FAO	Food and Agriculture Organization of the United Nations
H	Hemagglutinin
HA	Hemagglutinin antigen
HI	Hemagglutinin inhibition
HN	Hemagglutinin-neuraminidase

Executive Summary

Etiology and relevance

Bovine babesiosis, or redwater is a tick-borne disease caused by the intra-erythrocytic protozoan parasite *Babesia.* Only two *Babesia* species*, B. bovis and B. bigemina,* are important and widespread in tropical and subtropical countries. Although morphologically different, *B. bovis* being smaller than *B. bigemina*, the two species show variations that make it sometime difficult to discriminate among them.

Epidemiology and transmission

The lifecycle of the Babesias involves ticks and cattle. In ticks, their different forms and stages invade all tick's organs, including the ovaries, and insure passage to next generations of ticks. Babesia are maintained in cattle populations by asymptomatic carriers that have recovered from acute disease. *B. bovis* persists in cattle for years, and *B. bigemina* survives for a few months. Recrudescence of parasitemia can occur at irregular intervals. Babesia species are transmitted by ticks, which become infected when they ingest parasites in the blood of infected cattle.

The major vectors for *B. bigemina* and *B. bovis* are *Rhipicephalus microplus* (formerly *Boophilus microplus*) and *R. annulatus* (formerly *B. annulatus*). Other tick species, Rhipicephalus and others, can also transmit the two parasites. Bovine babesiosis can be found wherever the tick vectors exist, but it is most common in tropical and subtropical areas. *B. bovis* and *B. bigemina* are particularly important in Asia, Africa, Central and South America, parts of southern Europe, and Australia. *R. microplus* has recently been introduced to West Africa with cattle imported from Latin America and has been spreading at an alarming rate, with no measures being taken to curb the problem. There is however not yet enough evidence to support the possibility of subsequent spread of bovine babesiosis.

B. bovis and *B bigemina* affect cattle, which are the main reservoir hosts. They also affect water buffalo and African buffalo. Bovine Babesiosis is predominantly observed in adult cattle. Without treatment, mortality rates are very high (30% for *Babesia bigemina*, 70-80% for *B. bovis*). Calves can be infected *in utero*; however, this appears to require pathological changes in the placenta, and transplacental infection seems to be accidental and rare.

Very few studies exist on the different target countries and economic impact of bovine babesiosis; most of the information is on the overall impact of tick borne diseases (TBD). In East Africa for example, the importance and impact of bovine babesiosis is somehow obscured by that of Theileriosis. From the limited studies conducted to date, Minjauw et al. (2003), quoting a 1999 study, gave an estimated annual cost of bovine babesiosis in India of USD 57.2 million. In one of the few studies published in Africa, Kivaria's (2006) estimated the combined annual

direct costs associated with Babesiosis in Tanzania to be 45.8million USD, the third most important TBD after ECF and Anaplasmosis.

Reports on low prevalence and incidence for bovine babesiosis are considered to be underestimates, generally linked to the overwhelming focus on diseases like ECF in East and Southern Africa, the focus on ticks, which are a major and increasing problem in all countries covered in this report, poor diagnostic capacity and reporting that covers TBD as a group.

Clinical disease and diagnostic

Infections with *Babesia* spp. are characterised by a haemolytic syndrome which includes continuous fever, high parasitaemia, anaemia, icterus and often haemoglobinuria, which colours the urine dark brown and which gives the disease the common name of 'red water'. Infections associated with *B. bovis* are often acute or subacute, rapidly leading to death. Acute disease can cause nervous symptoms such as 'pedalling' movements and aggressive behaviour. In dairy cows, abortion and agalactia are early signs of infection.

Babesiosis can be diagnosed by identification of the parasites in blood or tissues, PCR, serology, or transmission experiments. In the case of *B. bovis*, a diagnosis of babesiosis can be confirmed by the presence of the parasites in Giemsa-stained blood smears. However, the presence of *B. bigemina* in a blood smear does not necessarily indicate babesiosis, as symptoms could be due to the resurgence of a chronic infection caused by another disease. Infected animals develop a life-long immunity against re-infection with the same species and some crossprotection is evident in *B*. *bigemina*-immune animals against subsequent *B*. *bovis* infections.

Control

Control of bovine babesiosis relies on tick control, chemotherapy/treatment, possibly the use of the use of tickresistant breeds of cattle and immunisation. The most widely available and effective drugs for the treatment of bovine babesiosis are diminazine aceturate and imidocarb dipropionate. The latter is an effective therapeutic agent for both anaplasmosis and babesiosis which also has prophylactic activity, and often is used to treat both diseases. Tick control, together with treatment, are the most widely used strategies in controlling bovine babesiosis in Asia and Africa.

Early bovine babesiosis immunization utilised infected blood collected from recovered animals—the so-called carrier donor system, and injected into young, relatively insusceptible calves, leading in most cases to a mild fever and subsequent immunity to what otherwise may have been a lethal natural challenge arising when animals were exposed to tick-infested pastures. The high variability in their infectivity and high virulence risk brought scientists in Australia to develop attenuated strains, through passage in splenectomised calves. The currently used blood vaccines are produced in splenectomised calves, which are inoculated with a stock of the

attenuated *B. bovis* and *B. bigemina* parasites, and bled during the acute phase of the reaction, to produce, with the collected blood, a standardized vaccine.

Most of the available live vaccines are produced in government-supported production facilities, notably in Australia, Argentina, South Africa, Israel and Uruguay. Of the target countries, only South Africa uses the bovine babesiosis vaccines, which are produced by the OVI and commercialised by OBP. The initial attenuated vaccines were chilled. But due to their short shelf life, a frozen form is now produced by OBP and many other countries, while Australia still produces the chilled form. Some manufacturers also have combination vaccines, including the two Babesia and Anaplasma. OBP produces the two babesia vaccines separately.

Since 1994, a vaccine with efficacy against the cattle tick, *R. microplus*, the major vector of bovine babesias, was developed, patented and marketed in Australia under the name TickGARD using the midgut glycoprotein antigen Bm86. A number of industrial consolidations produced unfavourable outcomes for TickGARD and it is no longer commercially available. A similar vaccine using basically the same antigen and oil adjuvanted was developed in Cuba in the 1990s, under the trade name Gavac, and is still being commercialised in North and Latin America. Review studies on the use of these two vaccines indicate that they have been effective in controlling *B. microplus*, resulting in subsequent decrease in acaricide usage and reductions in anaplasmosis and babesiosis. None of the tick vaccines is used in Africa and Asia.

The future of bovine baesiosis vaccines and vaccination

Of the countries targeted in the present monograph, only South Africa uses vaccination, with the only form currently available: the live attenuated bovine babesiosis vaccines produced in splenectomised animals. While they provide a good protection in immunized animals, their wide use is limited due to the complex and risky production process involving live animals, the requirement for strict cold chain and need for close veterinary or specialized supervision during administration. The poor results obtained with non-replicating vaccines and the discontinued efforts indicate that there may not be an alternative vaccine in the short to medium term. None of the Asian countries have been producing or using babesia live attenuated vaccines.

Options that could be considered in the meantime are:

1. Further improvement of the current live attenuated vaccine: aspects to be considered would include cell culture, freeze-drying, combination of both *B. bovis* and *B. bigemina* in Africa.

2. Combination with other TBD like Anaplasma, already practiced in Latin America could be considered given the common occurrence of these diseases.

3. Given the limited efficacy of inactivated vaccines, a vaccination program consisting of priming with inactivated vaccine and boosting with live one could be considered and evaluated as it may

address the safety problems of the live vaccine, while aiding in the buildup of a solid long lasting immunity.

4. There is a need to consider a broader use of the *R. microplus* tick vaccine in more regions in Africa and Asia. Although there is not yet evidence that the spread of the *R. microplus* tick in West Africa has corresponded to a worsening of Babesiosis, it will be critical to consider starting to deal with the problem before the parasite get established. Such vaccine could also be a way of addressing the worsening tick resistance problem in Africa.

Clinical disease overview

Bovine babesiosis or redwater is a tick-borne disease caused by the intra-erythrocytic protozoan parasite *Babesia*. Two species are economically important in tropical and subtropical regions of the world, including southern Africa: *Babesia bovis*, which causes Asiatic (European) redwater, and *Babesia bigemina*, which causes African redwater. *Babesia divergens* causes an economically important disease in the British Isles and northern Europe [9][25].

The present monograph will focus on Bovine babesiosis caused by *B. bovis* and *B*. *bigemina.* In the US situation it is referred to as Tick Fever, Cattle Fever or Texas fever. Another name is Piroplasmosis

Etiology

The parasite *Babesia* is named after Babes, the scientist who first described it in sheep and cattle in 1888 [9]. The genus *Babesia* belongs to the phylum Apicomplexa, class Sporozoasida, order Eucoccidiorida, suborder Piroplasmorina and family Babesiidae [2]. The *Babesia* spp. known to infect cattle in southern Africa, and their proven vectors, are listed in Table 1.

Parasite structure

Organisms belonging to the genus *Babesia* are pear-shaped and parasitize red blood cells of mammals and invade the internal organs of ticks. *Babesia bovis* is classically known as a 'small' *Babesia* measuring up to 2 µm in diameter, while *B. bigemina* is larger and can extend to the full diameter of an erythrocyte. However, the two species both show considerable morphological variation, making it difficult to identify one from the other. Large forms of *B. bovis* are quite common [9].

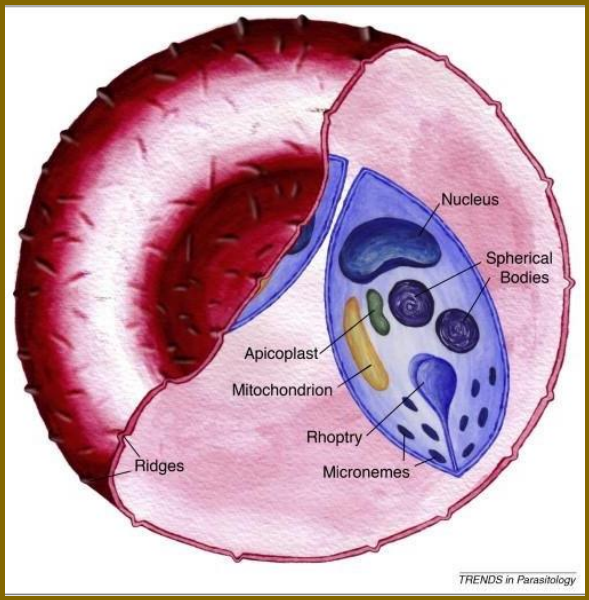

Figure 1: Schematic representation of a bovine Red Blood Cell parasitised by a mature form of *Babesia bovis*

Table 1: Bovine *Babesia* spp. and their vectors in southern Africa [9]

Babesia spp.	Vectors
Babesia bovis	*Rhipicephalus(Boophilus) microplus*
Babesia bigemina	*Rhipicephalus (B.)decoloratus* *Rhipicephalus microplus* *Rhipicephalus evertsi evertsi*
Babesia occultans	*Hyalomma marginatum rufipes*
Unnamed *Babesia* sp.	*Hyalomma truncatum*

Epidemiology

Susceptible animal species

Babesia bovis and B. bigemina have high degrees of host specificity: they are found in cattle, which are the main reservoir hosts. They also affect water buffalo (*Bubalus bubalis*) and African buffalo (*Syncerus caffer*).

In Africa, European, Sanga and Zebu breeds are all known to be susceptible, and all develop latent infections after recovery which persists for various lengths of time [9]. European breeds of cattle can retain *B. bovis* infections for life and remain infective for ticks for up to two years [5] while most cattle with a significant Zebu content lose the infection within two years. *Babesia bigemina* infections rarely persist for more than a year, regardless of the host, and infected cattle remain infective for ticks for only four to seven weeks.

African buffalo (*Syncerus caffer*) and Asiatic buffalo (*Bubalus bubalis*) can develop latent infections [9]. Antibodies and transitory infections have also been demonstrated in other animals, but there is little evidence to suggest that non familiarhosts are important reservoir hosts even though *R. e. evertsi* infected with *B. bigemina* have been collected off a sable antelope (*Hippotragus niger*) with clinical babesiosis.

Transmission and Vectors of Bovine Babesiosis

Babesia species are transmitted by ticks, which become infected when they ingest parasites in the blood of infected cattle. The major vectors for *B. bigemina* are *Rhipicephalus microplus* (formerly *Boophilus microplus*) and *R. annulatus*. *R. decoloratus*, *R. geigyi*, and *R. evertsi* can also transmit this species [5][9]. The major vectors for *B. bovis* are *R. microplus* and *R. annulatus*, but *R. geigyi* can also be a vector.

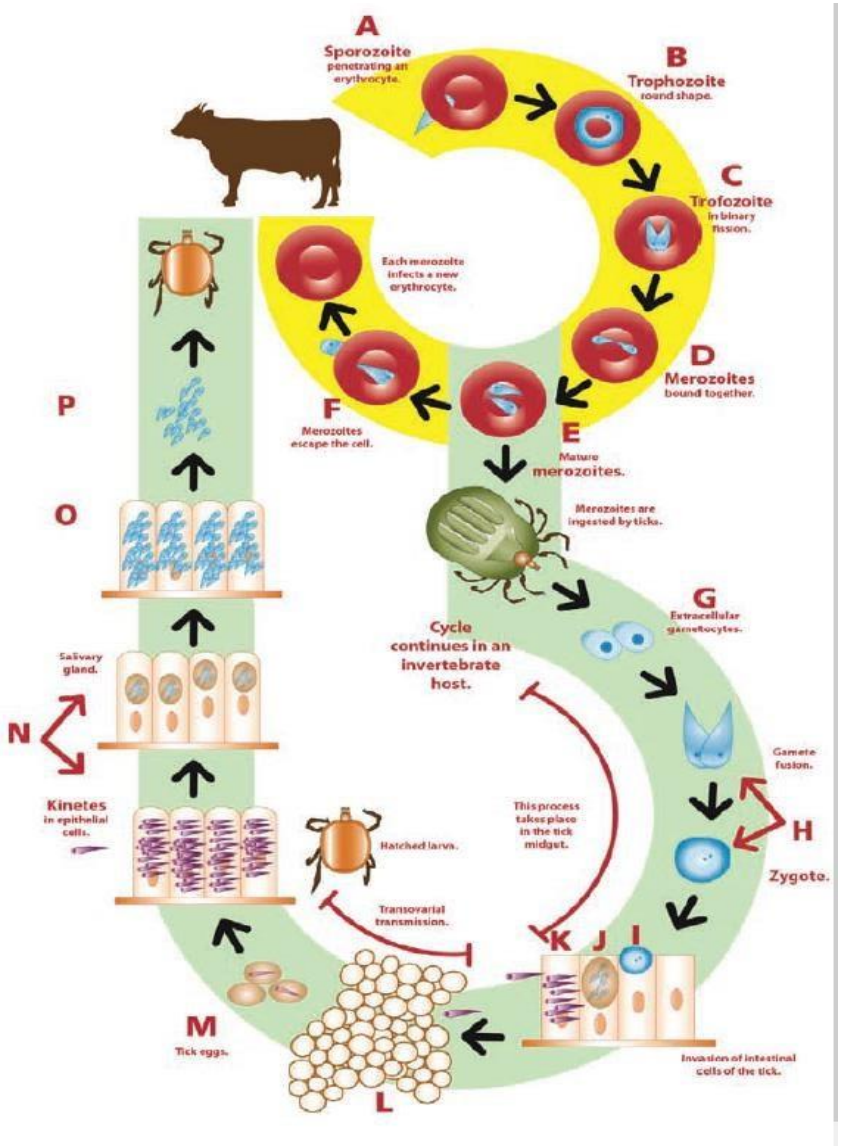

Figure 2: *Babesia bovis* life cycle (Mosqueda et al.; 2012)

Inside the tick, *Babesia* zygotes multiply as 'vermicules,' which invade many of the tick's organs including the ovaries; *Babesia* species are readily passed to the next generation of ticks in the egg. These parasites can sometimes be passed transovarially though several generations, although this varies with the species of *Babesia*

and the species of tick. *B. divergens* can survive in tick populations for at least 4 years even if cattle are not present. When an infected tick attaches to a new host, *Babesia* are stimulated to undergo their final maturation. *B. bovis* parasites usually become infective within 2-3 days after larval ticks attach, and can be transmitted by larvae. In *R. microplus*, *B. bovis* does not persist after the larval stage. In contrast, *B. bigemina* matures in approximately 9 days after a larval tick attaches, and it is only transmitted by nymphs and adults [5].

Babesia species can also be transmitted between animals by direct inoculation of blood. Biting flies and fomites contaminated by infected blood might act as mechanical vectors, although this method of transmission is thought to be of minor importance.

Babesia are maintained in cattle populations by asymptomatic carriers that have recovered from acute disease. *B. bovis* persists in cattle for years, and *B bigemina* survives for a few months. Recrudescence of parasitemia can occur at irregular intervals. Calves can be infected *in utero*; however, this appears to require pathological changes in the placenta, and transplacental infection seems to be accidental and rare. [9][24]

Inter-specific mating may occur between *B. microplus* and *B. decoloratus*, resulting in the production of sterile eggs and the creation of a 'hybrid zone' where populations of the two species overlap [5][9].

This zone may have prevented *B. microplus* from fully exploiting all the climatologically favorable areas in the subcontinent. Its width and stability are unknown, but it is likely to have a significant effect on the epidemiology of *B. bovis. Babesia bigemina*'s lack of vector specificity makes it unlikely that this species will be affected by competition between tick species.

One infected larval tick is sufficient to transmit *B. bovis*, but tick infection rates are usually low and the rate of transmission to cattle is therefore low. Tick infection rates with *B. bigemina* are higher (0,23 per cent in the Australian study) and therefore transmission rates of this species are also higher than those of *B. bovis.* As a result, *B. bigemina* infections are usually more prevalent in those herds where both species are present and less readily affected by factors such as climate or management, which reduce tick numbers [9].

Distribution

Bovine babesiosis can be found wherever the tick vectors exist, but it is most common in tropical and subtropical areas.

Based on CABI summary report (**http://www.cabi.org/isc/datasheet/91606)** *R. microplus* is originally from Asia and has been distributed, mainly with cattle, to all continents. The tick occurs in South and Central America, including Mexico, and is a major problem in Brazil. It occurs in much of southern Asia and also in China. The tick was introduced into East and South Africa from Madagascar, where it had originally arrived with cattle from southern Asia. In South Africa it is now established in scattered areas along the southern and eastern costs of the Western and Eastern Cape Provinces and of KwaZulu-Natal. *R. microplus* is also present in the coastal

regions of Mozambique, Kenya and Tanzania. The tick is spreading westwards in parts of southern (Zambia, Zimbabwe) and East Africa.

As discussed later in this document, *R. microplus* has since been introduced in West Africa and has been spreading at an alarming rate as far as Central Africa, with reports in Cameroon (1, Dr. Adakal personnal communication). But so far there is not enough evidence of a concurrent spread of Bovine babesiosis.

B. bovis and *B. bigemina* are particularly important in Asia, Africa, Central and South America, parts of southern Europe, and Australia. Although *B. bovis* is usually found in the same general geographic area as *B. bigemina*, slightly different groups of ticks spread these two species and some differences in their distribution can be seen. For example, *B. bigemina* is more widely distributed than *B. bovis* in Africa. *B. bigemina* and *B. bovis* and their vectors were formerly enzootic throughout much of the southern United States, but now are found only in a quarantine buffer zone along the Mexican border [24][26]. See Table 2.

Table 2: Bovine *Babesia* spp. and their vectors in southern Africa [9]

Organism	Animals affected	Geographical distribution
Babesia bigemina	Cattle, Zebu, Water buffalo Deer; Wild ruminants	Central and South America Australia Africa Southern Europe China
Babesia bovis	Cattle; Reindeer; Stag; Water buffalo, wild ruminants	Southern Europe Asia Africa America Australia

The distribution of the two pathogens in Africa is, as elsewhere aligned to that of the vectors, as shown in the Figures 3 and 4 below.

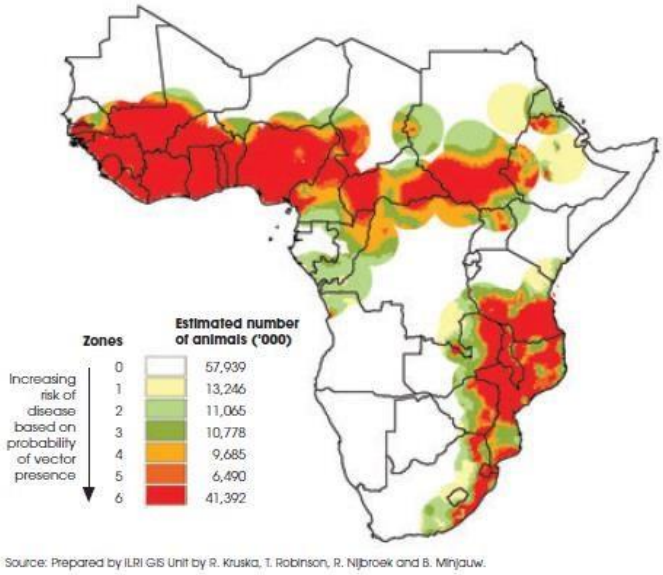

Figure 3: Map of Africa showing the distribution of *B. bovis*, related to the distribution of the vectors [19]

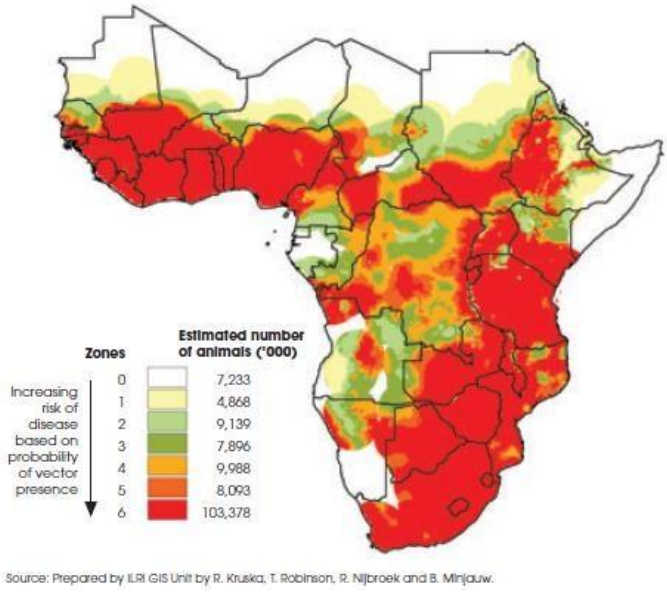

Figure 4: Map of Africa showing the distribution of *B. bigemina*, related to the distribution of the vectors [19]

Introduction and spread of *R. microplus* in West Africa:

- Over the past 10 years there have been an alarming spread and displacement of other ticks by *R. microplus* that is believed to have been introduced in the region from importation of cattle from Latin America [1]
- In contrast, the increased reporting on the spread of the tick has not corresponded to an increase in the number of outbreaks in the region (Personal communication Dr. Hassane Adakal), as illustrated by the OIE and AU-IBAR reports for the last 10 years.

Immunology

The mechanisms of immunity to babesial parasites are hypothesized to require innate as well as adaptive responses [7]. Innate immunity is non-specific and includes factors such as host–parasite specificity, genetic factors, age of the host and the response of host cells (such as the mononuclear phagocyte system and polymorphonuclear leukocytes). Most Babesia spp. are highly host specific and often splenectomy is needed to establish an infection in an unnatural host [5][6]. This is the basis for the production of the current live attenuated vaccine.

Two months old or younger calves born to previously unexposed cows are susceptible to infection and the effects of the disease, while offspring of immune mothers are resistant, presumably because of a passive transfer of immunity via the colostrum. After the age of two months a natural, non-specific, innate resistance, which persists for at least a further four to six months and is not dependent on the immune status of the cow, protects calves [9].

The mechanisms of immunity in babesiosis are complex and involve both antibody and cell-mediated components. Antibodies develop after infection, but not all are protective and attempts to correlate antibody levels with immunity have failed [7]. Live *B. bovis* inoculated into cattle induces antibodies that remain at high levels for at least six months. These antibodies are strain specific96 and are of the IgM and IgG1 isotype [9].

Specific and non-specific cell-mediated mechanisms involving T lymphocytes and natural killer (NK) cells appear to be involved in laboratory Babesia infections: intracellular death of parasites occurs which is not caused by antibodies and which is non-specific with other parasites, micro-organisms and even microbial extracts exerting the same effect on rapidly dividing Babesia organisms.

Macrophages are important for immunity to *B. bovis* because they act as antigen-presenting cells (APC) for T helper (Th) lymphocytes and remove parasitized erythrocytes by phagocytosis. Macrophages activated by interferon-gamma are thought to play a significant role in resistance to *B. bovis* by parasite suppression via secretory products released from macrophages [7].

Clinical Signs

In natural infections, incubation periods usually vary from 8 to 15 days. In acute manifestations, fever (>40°C) is usually present for several days before the onset of other clinical signs: inappetence, depression, weakness and reluctance to move. Haemoglubinuria is often present especially in *B. bigemina* infections (hence the common name "redwater"). Anaemia and icterus are especially obvious ion more protracted cases. Diarrhoea is common and pregnant cows may abort. Cerebral babesiosis, which occasionally develops in B. bovis infections, is manifested by hyperaesthesia, nystagmus, circling, head pressing, aggression, convulsions and paralysis; these signs may or may not accompany other signs of acute babesiosis [6][9][24]. Without treatment, mortality rates are very high (30% for *B. bigemina*, 70-80% for *B. bovis*) [19][24].

Necropsy of uncomplicated ("typical") cases of babesiosis is characterised by light red, watery blood and the mucous membranes and carcasses are paler than normal (these changes are due to anaemia). In many cases this pallor may be masked by icterus. Other findings include: enlarged and congested spleen, swollen friable and yellowish-brown liver, mottled appearance of the hepatic surface, distanded gall-bladder with viscous bile. The intestinal content is usually diminished (anorexia) and yellowish in colour (bile-stained). The kidneys are mildly to moderately swollen and dark reddish-brown in colour (haemoglobinuric nephrosis). The lungs are often oedematous, with foam present in the bronchi and trachea (probably due to agonal left heart failure). The heart is usually flabby and pale (degeneration, anaemia). The urine is discoloured and may be deep yellow to yellow-brown (bilirubinuria) or a clear port-wine colour (haemoglobinuria) [9].

Important to note that the above description of the macroscopic lesions applies not only to typical cases of babesiosis but to any disease in which significant erythrolysis occurs, hence the need for differential diagnosis taking into account other factors. Table 3 below summarises the major clinical signs of bovine babesiosis for *B. bigemina* and *B.bovis* .

Table 3: Clinical signs for *B. bovis* and *B. bigemina* (adapted from [6][9][26])

B. bovis	***B. bigemina***
High fever	Fever is less of a feature
Parasitaemia (percentage of infected erythrocytes) - maximum parasitaemia is often less than one per cent.	In *B. bigemina* parasitaemia often exceeds 10 per cent and may be as high as 30 per cent
Neurologic signs such as incoordination, teeth grinding and mania. Some cattle may be found on the ground with the involuntary movements of the legs. When the nervous symptoms of cerebral babesiosis develop, the outcome is almost always fatal.	Central nervous system (CNS) signs are uncommon Animals likely to separate from herd, be weak, depressed and reluctant to move

Dark coloured urine	Haemoglobinuria and anaemia Dark coloured urine
Anorexia	Anorexia
Lesions include enlarged soft and pulpy spleen, swollen liver, gall bladder distended with thick granular bile, congested dark-coloured kidneys and generalised anaemia and jaundice. Other organs may show congestion or petechial haemorrhages and occasionally there will be pulmonary oedema. Acute cases will show haemoglobinuria, but this may be absent in subacute or chronic cases. Clinical pathology centres on a haemolytic anaemia, which is characteristically macrocytic and hypochromic.	The pathogenic effects relate more directly to erythrocyte destruction. Haemoglobinuria is present earlier and more consistently than in *B. bovis* infections. Acutely affected cattle are usually not as severely affected as those with *B. bovis* infections. In some cases the disease can develop very rapidly with sudden and severe anaemia, jaundice and death, which may occur with little warning

Diagnosis

Differential Diagnosis

Babesiosis resembles other conditions that cause fever, and hemolytic anemia. The differential diagnosis includes anaplasmosis, trypanosomiasis, theileriosis, bacillary hemoglobinuria, leptospirosis, eperythrozoonosis, rapeseed poisoning and chronic copper poisoning. Rabies and other encephalitides may also be considerations in cattle with CNS signs [9][24]

Laboratory diagnosis

Laboratory diagnostics could be done either by blood smear, serology or using molecular techniques:

Blood smears: given the fact that babesia spp. parasites only develop in red blood cells, thin blood films made from capillary blood are preferred; although thick blood films may be more sensitive, species differentiation is more difficult. Blood of the general circulation may contain up to 20 times fewer *B. bovis* than capillary blood. In *B. bigemina* infections, parasitized cells are evenly distributed throughout the blood circulation. *B. bovis* parasitaemias are often low (<0.1%), even at the peak of the reaction, while B. bigemina parasites are usually more numerous and therefore easy to detect [9][26].

ELISAs have replaced the indirect fluorescent antibody (IFA) test as the most widely used test for the detection of antibodies to *Babesia spp*., because of processing efficiency and objectivity in interpretation of results. The IFA test has been used for detection of antibodies to *B. bigemina*, but serological cross-reactions make species

diagnosis difficult. The complement fixation (CF) test has also been used to detect antibodies against *B. bovis* and *B. bigemina* [26].

The OIE Manual reports that a number of other tests have been developed in recent years, including the slide ELISA, latex and card agglutination tests and an immunochromatographic test. These tests show acceptable levels of sensitivity and specificity for B. bovis and, in the case of the dot ELISA, also for *B. bigemina*. However, none of these tests appears to have been adopted for routine diagnostic use in laboratories other than those in which the original development and validation took place. Adaptability of these tests to routine diagnostic laboratories is therefore unknown [26].

The OIE also reports that a number of PCR techniques have been described for the detection and differentiation of Babesia species in carrier infections, as well as for differentiation of isolates of *B. bovis*. The application of the reverse line blot procedure, in which PCR products are hybridised to membranebound, species-specific oligonucleotide probes, to *Babesia* and, more recently, two quantitative PCR methods have enabled the simultaneous detection of multiple species, even in carrier state infections. However, current PCR assays generally do not lend themselves well to large-scale testing and at this time are unlikely to supplant serological tests as the method of choice for epidemiological studies. PCR assays are useful as confirmatory tests and in some cases for regulatory testing [9].

Incidence and Prevalence in Selected Countries

Global

Reports of the occurrence of Babesiosis specifically are not always made by many countries and often are grouped into reports of TBDs. Furthermore in East Africa Babesiosis as other TBD are masked by ECF clinically but also not often diagnosed. Most data are the result of research projects.

Table 4: Number of Bovine Babesiosis outbreaks reported to the OIE between 2005-2015 (Numbers given only for the target countries). Source: OIE.
http://www.oie.int/wahis_2/public/wahid.php/Diseaseinformation/statusdetail

Country	2005	2006	2007	2008	2009	2010	2011	2012	2013	2014	2015
Asia											
Bangladesh	0	0	0	0	0	0	0	0	0	0	0
India	36	26	45	48	130	128	131	121	181	274	0
Indonesia											
Myanmar (Burma)	0	0	1	1	6	8	27	23	46	22	0
Nepal	4	20	8	0	0	48	79	0	0	68	8
Vietnam	?	?	?	?	?	?	?	?	?	0	0

West Africa											
Burkina Faso	0	0	0	0	0	0	0	0	0	0	0
Ivory Coast	1	0	0	0	0	0	0	0	0	0	0
Mali	0	0	...	...	+..	...	...	...	...	...	...
Senegal	...	...	...	...	...	...	...	...	...	...	...
East Africa											
Ethiopia	+	0	0	+	+	+	+	+	+	0	0
Kenya	?	+..	?	+..	?	4	+..	+..	?	5	0
Rwanda		+..	+..	+..	+..	+..	+..	+	+..	...	...
Tanzania	10	29	184	56	33	36	27	12	18	20	7
Uganda	+..	+..	+..	+..	+..	+..	+..	+..	+..	+..	
Southern Africa											
Madagascar	0	0	0	+	+	4	10	8	13	7	0
Malawi	3	2	5	4	3	4	2	3	1	0	0
Mozambique	6	19	4	2	3	6	1	2	3	15	0
South Africa	195	97	73	85	73	91	59	37	16	28	0
Zambia	**0**	38	35	64	130	108	0	38	78	74	0

- No information, + Present but quantitative data not known, ? Disease suspected

2- AU-IBAR: The number of outbreaks reported to AU-IBAR is included in the Pan African Animal Resources Year Book. (http://www.au-ibar.org/pan-african-animal-resources-yearbook?showall=&limitstart=) and can be seen for the countries of interest in Table 5 below.

Table 5: Number of Bovine Babesiosis outbreaks reported to the AU-IBAR from 2005 to 2015 (numbers given only for the target countries). Source: AU-IBAR Year Books.

Country	2005*	2006**	2007	2008	2009	2010	2011	2012	2013	2014	2015
West Africa											
Burkina Faso											
Ivory Coast											
Mali											
Senegal				1							
East Africa											
Ethiopia				9			1				
Kenya			5	2	18 cases	64 cases	26	24	2	5	
Rwanda											
Tanzania		151	210	591 cases	148 cases		25	12	19	22	
Uganda				7	11	4					
Southern Africa											
Madagascar			1	68 cases	193 cases		3				
Malawi			5					3			
Mozambique		5	6	2	4	7	2	2	6	2	

South Africa		90	85	87	42	119		89	40	42	
Zambia			36	221 cases	22	571 cases	43	24	67	92	

*AU-IBAR didn't start yet producing data for bovine babesiosis
**No individual country report available; 7 countries reported to a total of 334 outbreaks

Regional

ASIA

Bangladesh

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2014	Rangpur district	Cattle	400	Gangachara: 1.3% Pirgachaupazilas: 1.7%	Rahman et al, 2015
2013-2014	Sirajgang	Cattle	395	2.27%	Al Mahumd et al, 2015
2011-2012	Sylhet district	Cattle	100	16% *B. bigemina*	Chandra Nath et al, 2013
2009-2010	Chittagong	Cattle	Crossbred: 216 Indigenous: 432	Crossbred: 9.25% Indigenous: 7.17%	Alim et al, 2012
2004	Sirajgong	Clinically suspected cattle	60	3.3% *B. bigemina*	Chowdhury et al, 2006

India

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2015	Central plain zone of Punjab	Cattle	204	2.45% *B. bigemina*	Bhat et al, 2015
2013	Three states of North-eastern India	Cattle suspected for haemoprotoxoan	333	3.60% *B. bigemina*	Laha et al, 2015
2013	National Research Centre on Yak,	Yak	94	5.32% *B. bigemina*	Saravanan et al, 2013
	Dirang, Arunachal Pradesh				
2012	Southern Punjab	Cattle and buffaloes	Cattle: 105 Buffaloes: 39	18.75%	Zulfiqar et al, 2012
2012	Ludhiana, Punjab	Cattle	703	1.56% *B. bigemina*	Singh et al, 2012
2011	9 districts Punjab	411 dairy animals	Cattle: 386 Buffaloes: 25	*B. bigemina*: 2.43% *B. bigemina + T. evansi*: 3.41%	Sharma et al, 2013
2011	Northern Kerala	Crossbred cattle	150	0.6% *B. bigemina*	Nair et al, 2011
2009	Endemic zones of Uttar Pradesh and Punjab	Cattle and buffaloes	Cattle: 180 Buffaloes: 120	56.11% SELISA 23.33% IFAT *B. bigemina*	Singh et al, 2009

Indonesia

No data was found for prevalence of babesiosis.

Myanmar

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2015	Nation wide	Cattle	713	*B. bigemina:* 9.8% *B. bovis:* 17.1%	Bawm et al, 2016

Nepal

No data was found for prevalence of babesiosis.

Vietnam

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2015	Thur Thien Hue province	Cattle and water buffaloes	Cattle: 258 Buffaloes: 49	Cattle: 8.9% W Buffaloes: 32.65% *B. bovis*	Yokoyama et al, 2015
2014	Central region	Cattle and water Buffaloes	137	PCR: *B. bovis:* 21.3% *B. bigemina*: 16.0% ELISA: *B. bovis*: 37.2% *B. bigemina*: 9.3% IFAT *B. bovis:* 27.9% *B. bigemina:* 18.6%	Li et al, 2014

2010-2011	Hue and Hanoi provinces	Cattle, buffalo, sheep and goats	Cattle: 201 Buffalo: 43 Sheep: 51 Goats: 127	Cattle: *B. bovis*: 8.9% *B. bigemina*: 3.5% Buffalo: *B. bovis*: 9.3% *B. bigemina*: 0 Sheep: 0 for both Goats: *B. bovis*: 0 *B. bigemina*: 0.8%	Sivakumar et al, 2013
2008	Around Hanoi	Dairy cattle	239	54% *B. bigemina*	Geurden et al, 2008

AFRICA

Burkina Faso

Year	Area	Species of animal	No. of samples tested	% positive	Reference
1993-1996	Baoura and Kourouma	Cattle	799	*B. bigemina* isolated from 1 dead calf	Ganaba et al, 2000

Ethiopia

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2015	Around Jimma Town South Western Ethiopia	Cattle	400	23%	Lemma et al, 2015
2014-2015	Assosa District, Beishangul Gumuz Regional State	Cattle	402 blood samples	1.5% (blood semar)	Wodajnew et al, 2015
2014	Northern Ethipia	Cattle, sheep and goats	525	0	Gebrekidan et al, 2014
2013-2014	Teltele District, North West Borena Zone, Southern Ethiopia	Cattle	384 blood samples	Overall: 16.9% Fulotole: 9.4% Hatuse: 13.6% Kulcha: 18.2% Billa kebele: 27.85%	Hamsho et al, 2015
2010-2011	around Debre-Zeit, Central Ethiopia	Cattle	384 blood smears	0.9%	Sebele et al 2015
2002	Ghibe Valley	Indigenous cattle	60	Indirect ELISA 87.1% *B. bigemina*	Feleke et al, 2008

Ivory Coast

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2010	Bingerville area	Ticks from 36 cattle	120 Rhipicephalus microplus ticks	Giemsa: 1.73%	Toure et al, 2010

Kenya

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2011	Ngong and Machakos	Cattle	154 Ngong 38 Machakos	Ngong: *B. bigemina*: 42.2% *B. bovis:* 12.3% Machakos: *B. bovis:* 23.7% *B. bigemina:* 13.2%	Moumoni et al, 2015
2010	Mbeere District	Cattle	440 in 80 farms	19% *B. bigemina*	Gachochi et al, 2010
2007-2009	Western Kenya	Indigenous calves	548	36% *B. bigemina*	Kiara et al, 2014
2007-2009	Western Kenya	Indigenous cattle (dams)	548	48.9%	Toye et al, 2013
2007-2009	Machakos	Cattle	634 zebu 15 crossbreds	563 samples tested: 38% *B. bigemina*	Wesonga et al, 2010
1996-1998	Western highlands	Cattle	Rural: 600 Peri-urban: 160	Rural: 0.42% Peri-urban: 1.45%	Okuthe, 2006

Madagascar

No recent information on prevalence of babesiosis was found.

Malawi

No recent information on prevalence of babesiosis was found.

Mali

Year	Area	Species of animal	No. of samples tested	% positive	Reference
1984	Bamako	Cattle		*B. bovis:* 38%% *B. bigemina*: 54% IFAT	Miller et al, 1984

Mozambique

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2010	Maputo Province	Healthy cattle	477	PCR: *B. bigemina:* 30-89% *B. bovis*: 27-83% Reverse line blot: *B. bigemina:* 0-17 *B. bovis:* 0	Martins et al, 2010
2010	Maputo, Gaza and Inhambane		809	*B.bovis:* 78.8% *B. bigemina: 76%*	Tembue et al, 2011
2008	Near Maputo city	Cattle	117	PCR *B. bigemina:* 90% *B. bovis*: 82% Mixed: 56%	Martins et al, 2008
2005	Tete province	Cattle	478	*B. bovis*: 39%	Alfredo et al, 2005

Rwanda

No recent information on prevalence of babesiosis was found.

Senegal

No recent information on prevalence of babesiosis was found.

South Africa

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2010-2012	All nine provinces of South Africa	Healthy cattle	430	B. bigemina: 64.7 B. bovis: 35.1 For details, see table below	Mtshali and Mtshali, 2013
2011	Eight provinces	Healthy cattle	719	*B. bovis:* 35.3% *B. bigemina:* 39.7%	Terkawi et al, 2011
2009-2010	Gauteng province	Healthy cattle	268	*B. bovis*: 35.5% *B. bigemina:* 76.1%	Mtshali et al, 2013
2010	Northeastern edge of the Hluhluwe-iMfolozi Park	Nguni cattle	60	*B. bovis:* 303% *B. bigemina:* 1.7%	Yusufmia et al, 2010
2007-2008	Magwiji, Ukhahlamba district	Cattle	200	*B. bovis:* 45% *B. bigemina:* 46%	Munyaradzi et al, 2010
1999-2000	Limpopo	Cattle	30 communal dip tanks, and 5 commercial farms	*B. bovis:* 1999: 63.3% 2000: 62.4%	Tonnesen et al, 2006

				B. bigemina: 1999: 56.1% 2000: 49.3%	
1999-2000	Free State	Cattle	386	*B. bigemina:* (IFA) 94%	Mtshali et al, 2004

Province	Total number of samples	*Babesia bigemina*		*Babesia bovis*		Mixed infection	
		No. positive	Percentage	No. positive	Percentage	No. positive	Percentage
Mpumalanga	48	32	66.7	21	43.8	15	31.3
KwaZulu-Natal	52	44	84.6	33	63.5	28	53.9
Limpopo	47	39	83.0	11	23.4	11	23.4
North West	58	41	70.7	38	65.5	24	41.4
Gauteng	30	25	83.3	19	63.3	18	60.0
Free State	64	56	87.5	15	23.4	14	21.9
Eastern Cape	60	21	35.0	7	11.7	4	6.7
Northern Cape	45	2	4.4	2	4.4	0	0
Western Cape	26	18	69.2	5	19.2	3	11.5
Total	**430**	**278**	**64.7**	**151**	**35.1**	**117**	**27.2**

Tanzania

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2013	Monduli district	Indigenous cattle	295	*B. bovis:* 1.7%	Haji et al, 2014
1999	Tanga and Iringa	Dairy cattle	1,395	*B. bovis:* Tanga: 6% Iringa: 12%	Swai et al, 2004
1999	2 contrasting regions of Tanga (tropical	Smallholder dairy cattle	1329; ELISA test	Iringa 43% Tanga 27%	Swai et al, 2007

	coast) and Iringa (tropical highlands)			Overall 34.9%	

Uganda

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2014	Kiruhura district, in two sub-counties near Lake Mburo National Park in South-western Uganda	Ankole Long-horned cattle and European crossbred cattle	130	26.9%	Schischkle, 2015
2013-2014	Central and Western Uganda	Bovine samples	295	*B. bigemina:* 6.7%	Kasozi et al, 2014
2008	Kashaari county	Cattle	363	0.6%	Muhanguzi et al, 2010
2010	National Parks	Cape buffalo		No Babesia found	Oura et al, 2011

Zambia

Year	Area	Species of animal	No. of samples tested	% positive	Reference
2009		Sanga cattle	71	*B. bigemina:* 22.5%	Yamada et al, 2009

Economic and Social Impacts at Global and Regional Levels, and in Selected Countries

As for other TBD, in Eastern and Southern Africa, as well as in India, the importance and impact of Bovine babesiosis is somehow obscured by that of Theileriosis. From the limited studies conducted to date, Minjauw et al. [19], quoting a 1999 study, give an estimated annual cost of Bovine Babesiosis in India of USD 57.2 million.

Kivaria's [27] study estimated the combined annual direct costs associated with Babesiosis in Tanzania to be 45.8million USD, the third most important TBD after ECF and Anaplasmosis. The main components are given in the table below.

Table 6: Combined annual direct cost associated to Babesiosis in Tanzania. [27]

Item	000 000 USD
Control	
Acaricides	5.04
Chemotherapy	8.60
Production losses	
Mortality	21.54
Milk loss	1.58
Live weight	9.06
Total	**45.82**

Makala et al, reported in 2003 that an underestimated number of 200,000 cattle are exposed to bovine babesiosis in Zambia. With the high number of naïve *B. taurus* being introduced in many regions, high mortalities are recorded [28].

According to Bock [5] citing a model developed by McLeod & Kristjanson (1999) in 1999, estimated costs incurred due to losses and control of babesiosis and anaplasmosis in Kenya, Zimbabwe, Tanzania, South Africa, China, India, Indonesia and Philippines cost 5.1, 5.4, 6.8, 21.6, 19.4, 57.2, 3.1 and 0.6 million US dollars annually, respectively

He states that costs due to babesiosis are incurred not only from mortality, ill-thrift, abortions, loss of milk/meat production and draft power and from control measures (such as acaricide treatments, purchase of vaccines and therapeutics), but also through its impact on international cattle trade.

Disease Prevention and Control Methods

The experience in the US has shown that Babesiosis can be eradicated by eliminating the host tick(s). This was accomplished by treating all cattle every 2 to 3 weeks with acaricides (APHIS 2010). In countries where eradication is not feasible, tick control can reduce the incidence of disease. The development of resistance to acaricides can be a concern. Environmental modification can also destroy tick habitats, but in some cases this may be difficult and/ or ecologically undesirable.

Four methods are available for the control of heartwater: tick control, antibiotic treatment of clinical cases, prophylactic use of antibiotics, and immunization. The combination of these 4 methods is also an option. Tick control through tick vaccination has been used in Australia [14].

An important element in the control of bovine babesiosis in endemic regions is the concept of endemic stability.

Endemic stability

In areas where there is a continuous inoculation of cattle with *Babesia spp*. by infected ticks, calves are likely to be in contact with the parasite during the first 10–12 months of life, when typically they do not show any clinical manifestations. Babesia parasites are able to establish persistent infections in these animals that thus develop into parasite carriers with strong acquired immunity and resistance to disease [9]. This situation is described as enzootic stability; the first description stated that if 75% of the animals in a herd have been exposed to *B*. *bovis* before 9 months of age, as indicated by specific antibody titres, the chance of observing clinical cases is very low. Although natural endemic stability is in principle an ideal condition where no control measures are needed, this situation is rare and, when it does exist, it can be easily broken by variations in climate, host genotypes and management strategies. In addition, artificially maintaining endemic stability by yearly manipulating transmission rates in each herd is highly impractical.

Importantly, it has been argued that the threshold of 75% exposed cattle as a means to predict the appearance of clinical cases should be taken with caution when extrapolated to different regions and host-tick-pathogen systems from those on which the model was based [9]. In regions of enzootic instability, or when cattle are relocated from tick-free to tick-infested regions, prophylactic immunization has proved an effective method to prevent the occurrence of babesiosis outbreaks [5].

Treatment (Control)

Vector control

Eradication of the tick vectors (the so-called minimum disease situation) is the most desirable, permanent solution to the problem but is rarely considered practical or economical. The alternative approach, allowing natural endemic stability to develop by practicing limited or no tick control, is similarly unrealistic in areas where *R. appendiculatus* and *Amblyomma* spp. are well established [21]. These regions are also endemic for other *Rhipicephalus (Boophilus)* spp. and essential control of other tick species will inevitably affect the epidemiology of redwater. In the long-term, this approach can be achieved by integrating the strategic use of acaricides, the application of vaccines in endemically unstable conditions and the use of tick-resistant breeds of cattle [9][21]

Tick vaccine

A subunit vaccine against R *microplus* has been developed and commercialized in Australia since 1994, based on the Bm86 antigen, and called TickGARD. A similar vaccine, possibly based on the Australian vaccine, has been produced in Cuba and commercialized also in parts of South America. The vaccine has been the result of more than 12 years of research and field trials with 18,000 cattle.

While the vaccine is mostly used in an integrated way resulting in reduced reliance on pesticide over years of use, Lighthowlers [14] reports that there are observations and studies that have shown:

- Reduction in incidence of anaplasmosis and babesiosis in Cuba in certain areas (Cuba produces and use the tick vaccine),
- No transmission of *B*. *bigemina* and
- Reduced effect of *B*. *bovis* transmitted by *R*. *annulatus* on TickGARD-vaccinated cattle

However, to date, the use of tick vaccine is still limited even in countries where they are produced, except for some Latin American countries where the Cuban vaccine is exported. The Australian technology has not been exported due to ownership and IP issues.

Treatment

A number of drugs have been used to date and shown ability to cure infection [9]. Recovery is generally achieved if specific treatment is given early in the course of the infection. If treatment is delayed, however, supportive therapy may be essential if the animal is to survive. Blood transfusions may be indicated in cattle with heavy parasitaemias and low PCVs (<0.10); histo-incompatibility seldom occurs at the first transfusion. In acute *B. bovis* infections, use of antioxidants such as vitamin E, and high doses of corticosteroids may help to offset the

hypotensive and hypercoagulable state of the animal. In cases of cerebral babesiosis, intravenous use of hypertonic solutions of mannitol or glucose may provide temporary relief.

Drugs that are successfully used include:

- Diamidine derivatives: diminazenes (such as Berenil, Trypazen), Amicarbalides, Imidocarb and Phenamidine
- Quinoline derivatives: quinuronium sulphates (Babesan, Acaprin etc.)
- Acridine derivatives: Euflavine, Trypan blue
- Antibiotics: Tetracycline

Vaccination following the use of chemotherapeutic agents is probably the most frequent cause of vaccine failures. The immunity conferred by the live vaccines depends upon the establishment of the infection in the host as well as the degree of antigenic stimulation. Chemotherapeutic drugs administered before or during babesiosis vaccination will interfere with the multiplication of the parasites and thus the development of immunity. The practical implication of this becomes clear when the vaccine is used after the prophylactic treatment of cattle with imidocarb, and to a lesser extent, diminazene. The residual effect of these drugs will protect the cattle against natural infection for a limited period of time [10]

This has necessitated the changing of recommended waiting periods before vaccinating with the frozen vaccine in the case of *B. bovis* from 8 to 12 weeks and for *B. bigemina* from 16 to 24 weeks [10]

Chemoprophylaxis

Chemoprophylaxis as a method of short-term protection against babesiosis is often used under particular instances such as the temporary residence of susceptible animals in an infected area (for example agricultural shows), when pregnant cows are at risk and during disease outbreaks.

Imidocarb and diminazene are the only babesiacides with useful prophylactic properties for the short-term control or prevention of babesiosis. Treatment with imidocarb (3 mg/kg) will prevent overt *B*. *bovis* infections for at least four weeks and *B*. *bigemina* infections for at least eight weeks. Diminazene (3,5 mg/kg) will protect cattle against the two diseases for one and two weeks, respectively. Unfortunately, the prophylactic use of imidocarb may interfere with the development of immunity following vaccinations because the residual effect of the drug may eliminate or suppress the infection [9][25].

Prophylaxis (Prevention)

Immunisation

Prophylactic immunization has proved most effective to reduce losses caused by bovine babesiosis. Live, attenuated strains of *B. bovis* or *B. bigemina* are used to vaccinate cattle in some countries. These vaccines have safety issues including the potential for virulence in adult animals, possible contamination with other pathogens, and hypersensitivity reactions to blood proteins [26]. They are best used in animals less than a year of age to minimize the chance of disease. In some cases, vaccination of older cattle is necessary (e.g., if susceptible cattle are moved into an endemic area). Older animals should be monitored closely after vaccination, and treated if clinical signs develop. In some countries, animals may be vaccinated in the face of an outbreak (OIE Manual). The use of genetically resistant cattle such as *B. indicus* can also decrease the incidence of disease. Natural endemic stability is unreliable as the sole control strategy, as it can be affected by climate, host factors and management [9]. See more details in Section 6.

Control of outbreaks

Procedures to be followed during an outbreak will depend largely on the number and manageability of the animals concerned, and the availability and cost of labor, drugs, vaccine and acaricides. One or more of the following actions can be taken to limit losses:

- Treat sick animals and separate them, if possible, from the rest of the herd.
- Have the diagnosis confirmed at a reputable laboratory.
- Treat unaffected cattle for ticks to prevent exposure.
- Consider immediate vaccination of all unaffected cattle.
- Consider use of a prophylactic treatment programme as mentioned above.

Disease situation and government policies by country:

Tables 7 and 8 below have been completed with the information received so far from the questionnaires sent to the DG and DVS. This information will be updated and completed once the results from the different countries are received.

Table 7 covers the disease situation (if it is notifiable or not), the presence of official surveillance and/or control programs, and the treatment situation. Table 7 refers to vaccination.

The definitions that were given to the respondents are:

[1]Surveillance: is the systematic ongoing collection, collation and analysis of data and the timely dissemination of information to those who need to know so that action can be taken.

[2]Control: a program which is approved, and managed or supervised by the Veterinary Authority of a country for the purpose of controlling a vector, pathogen or disease by specific measures applied throughout that country, or within a zone or compartment of that country.

Table 7: Official status, official programs for Babesiosis in the countries of interest.

Country	Notifiable (yes/no)	Official surveillance[1] program (yes/no) (if yes, active or passive)	Official control[2] program (yes/no)	Treatment (Chemotherapy)	
				Treatment authorised (yes/no)	Frequently practiced (yes/no)
ASIA					
Bangladesh	Yes	No	No	Yes	Yes
India					
Indonesia					
Myanmar (Burma)	No	No	No	No	Yes
Nepal	No	Yes, passive	No	Yes	Yes
Vietnam	No	No	No	-	-
AFRICA					
Burkina Faso					
Côte d'Ivoire (Ivory Coast)	Yes	Yes, passive but active if outbreak	NO	Yes	If the animals are sick
Ethiopia					

Kenya	Yes	Yes, passive	No	Yes	Yes
Madagascar					
Malawi	No	No	Yes	Yes	Yes
Mali	N/A	N/A	N/A	N/A	N/A
Mozambique					
Rwanda	-	-	-	-	-
Senegal					
South Africa					
Tanzania	No	Yes, passive	Yes	Yes	Yes
Uganda	No	No	No	N/A	N/A
Zambia					

Table 8: Vaccination for Babesiosis in the countries of interest.

Country	Vaccination			
	Compulsory vaccination (yes/no)	Who pays for the vaccine (Government, farmers, combination, others-specify)	Who delivers the vaccine (official, private vaccinators or both)	Species vaccinated (cattle, sheep, goats, pigs, poultry)
ASIA				
Bangladesh	No	-	-	-
India				

Indonesia				
Myanmar (Burma)	No	-	-	-
Nepal	No	N/A	N/A	N/A
Vietnam	No	-	-	-
AFRICA				
Burkina Faso				
Côte d'Ivoire (Ivory Coast)	No	-	-	-
Ethiopia				
Kenya	No	Farmers	Both	Cattle
Madagascar				
Malawi	No	N/A	N/A	N/A
Mali	N/A	N/A	N/A	N/A
Mozambique				
Rwanda	-	-	-	-
Senegal				
South Africa				
Tanzania	No	N/A	N/A	N/A
Uganda	No	Never vaccinated	N/A	N/A
Zambia				

Vaccines Available

The development of vaccines against bovine babesiosis was prompted by early observations indicating that cows that recovered from natural *Babesia spp*. infections developed long-lasting immunity; and inoculation of their blood into susceptible cattle resulted in a less virulent form of the disease. Thus, the first vaccine formulations consisted of blood from donor bovines that had recovered from infection [9][10].

A breakthrough in the development of bovine babesiosis vaccines was achieved by Australian researchers, who observed that rapid successive blood passages of *B*. *bovis* between splenectomized calves resulted in progressive virulence decrease, with diminished post-vaccination changes in body temperature and haematocrit. Later, attenuation of *B*. *bigemina* was also achieved, but in this case the procedure involved slow successive passages among spleen-intact calves [14].

Attenuation also leads to the waning of nervous symptoms in the case of *B. bovis* and is sometimes, but not always, associated with a loss of tick transmissibility [15]. Since the spleen is important in the trapping and destruction of infected erythrocytes, the use of splenectomized bovines yields adequately high parasitaemias in the case of B. bovis [5]. Current vaccines against *B*. *bovis* and *B*. *bigemina* are based on these attenuation procedures.

All currently used vaccines are live attenuated and contain attenuated Australian strains of *Babesia*, mainly *B. bovis* and *B. bigemina* (and *Anaplasma* for some) in a multivalent or monovalent vaccines, and are produced in government-supported production facilities as a service to the livestock industries, in particular in Australia, USA, Israel, a number of Latin American countries and South Africa (Table 9). Some other countries possess the ability to produce vaccine on a smaller scale [6][26].

Various ways of attenuating *Babesia* spp. have been reported. The most reliable method of reducing the virulence of *B. bovis* involves rapid passage of the strain through susceptible splenectomised calves. Attenuation is not guaranteed, but usually follows after 8 to 20 calf passages [6]. The virulence of *B. bigemina* decreases during prolonged residence of the parasite in latently infected animals. This feature has been used to obtain avirulent strains by infecting calves, splenectomising them 6–12 weeks after inoculation and then using the ensuing relapse parasites to repeat the procedure [6].

Calves to be used as vaccine donors should be splenectomised to allow maximum yield of parasites for production of vaccine. This is easier in calves less than 3 months of age and is best performed under general anaesthesia [26]. Donor cattle for babesiosis vaccine are bred and maintained tick-free. They should routinely be examined for agents of all blood-borne infections prevalent in the country, including *Babesia, Anaplasma, Theileria*, *Eperythrozoon* and *Trypanosoma*. The absence of other infective agents endemic in the country, viral, bacterial or other, should also be confirmed.

For the frozen South African vaccine production, the splenectomised calves are infected with attenuated *B. bigemina* or *B. bovis* vaccine strains, and their heavily infected blood is collected in an anticoagulant at the peak of the reactions. The infection is quantified and adjusted by dilution with uninfected whole blood to ensure a standardized number of parasites per vaccine dose. To this is added an equal volume of phosphate buffered saline diluent containing 20% DMSO as cryoprotectant. Monovalent *B. bigemina* and *B. bovis* are produced in quantities of five doses each, deep frozen in liquid nitrogen and issued on demand [10].

The five doses vaccine vials are then stored in liquid nitrogen, while quality control procedures are conducted. The released vaccine is then distributed in dry-ice to users in the country.

Since 1998 there has been an increased preference of the ultra-frozen vaccines over the chilled vaccine [10]. The advantages of the frozen vaccines include: Lower production cost,

- Prolonged shelf life: chilled live vaccines have limited shelf-life, lasting between 4 and 7 days at 4 °C [5].
- On demand availability: vaccine can be stored in liquid nitrogen for up to 18 months; this increase cost efficiency
- The pre-release safety and efficacy testing can be conducted while the vaccine is kept frozen

Live Babesia vaccines are recommended to be used in calves 4 to 10-month-old that generally show good tolerance, though a transient clinical response to vaccination can sometimes take place [25]. Adult animals, on the other hand, can develop acute babesiosis upon vaccination, for which daily monitoring for up to 21 days is suggested and babesiacide treatment is often needed [23][10]. It was observed in South Africa that, after chemosterilization of infections, sterile immunity to *B. bigemina* lasted for only 16 months, without further boosting of immunity from tick-acquired infections, while immunity to *B. bovis* lasted for over 3 years [10]. Thus, complete tick control after vaccination is discouraged, so that natural infections through tick bites can aid in the acquisition of a long-term protected status [5][10].

The main features of the live Babesia vaccines are as follows [9]:

- Protective immunity develops in four to six weeks.
- The immunity lasts for several years in the case of *B. bovis*, but in the absence of natural challenge, it may break down in the case of *B. bigemina.* However, the latter does not seem to pose a problem under endemic conditions and revaccination is therefore rarely advocated.

- Babesiosis vaccines can be given at the same time as anaplasmosis and other vaccines, except heartwater vaccine.
- Attenuated strains of *B. bovis* and *B. bigemina* impart a high degree of immunity to other strains, even in other countries and continents.
- Factors influencing the development of protective immunity include potency of the vaccine (viability), development of 'breakthrough' field isolates, poor immunogenicity of the vaccine strain and poor responsiveness of the vaccinated animals.

Table 9 below provides a summary of bovine babesiosis vaccines produced around the world. It is important to note that in many cases Bovine Babesiosis vaccines are combined with Anaplasmosis vaccines. Although several studies have been published over the years on other forms of Bovine babesiosis vaccine, none have been commercialized.

Table 9: Bovine babesiosis vaccine manufacturers around the world [11]

Country	Vaccine name	Antigen	storage	Reference or contact
Argentina	VACUNA CONTRA LA BABESIOSIS Y LA ANAPLASMOSIS/INTA-Rafaela	*A. centrale , B. bigemina, B bovis*	R	
	BIOJAJA/Laboratorio Litoral Biológico	*A. centrale , B. bigemina, B bovis*	U-F	http://www.veterinariargentina.com/revista/2013/08/tristeza-bovinahttp://www.veterinariargentina.com/revista/2013/08/tristeza-bovina-vacuna-producida-en-el-chaco-argentina/vacuna-producida-en-el-chaco-argentina/ http://www.senasa.gov.ar/contenido.php?to=n&in=1454&io=12690
	VACUNA CONTRA LA BABESIOSIS Y LA ANAPLASMOSIS / INTA-Mercedes	*A. centrale , B. bigemina, B bovis*	R	http://inta.gob.ar/documentos/vacunas-para-la-babesiosis-y-anaplasmosishttp://inta.gob.ar/documentos/vacunas-para-la-babesiosis-y-anaplasmosis-tristeza-.-noticias-y-comentarios-503/at_multi_download/file/INTA_Vacuna Babesiosis y Anaplasmosis _Not y com 504.pdftristeza-.-noticias-y-comentarios-503/at_multi_download/file/ INTA_Vacuna%20Babesiosis%20y%20Anaplasmosis%20_Not%20y%20com%20504.pdf
Colombia	Anabasan ANABASAN®/ Limor de Colombia SA	*A. centrale , B. bigemina, B bovis*	U-F	http://corpomail.corpoica.org.co/BACFILES/BACDIGITAL/45107//s2dF1DFA3 755B2E7DB298E703DD8F72FAF0_1.pdf http://www.limorcolombia.com/biotecnologia.html
USA	Anavac PHL Associates			Anaplasma (Attenuated)
Australia	Chilled Tick Fever Vaccine Department of Primary Industries and Fisheries, Tick Fever Centre (DAFF-TFC)	*A. centrale , B. bigemina, B bovis*	R	http://www.daf.qld.gov.au/__data/assets/pdf_file/0008/61388/Tick-Feverhttp://www.daf.qld.gov.au/__data/assets/pdf_file/0008/61388/Tick-Fever-A2- Trivalent-Tick-Fever-Vaccine-Specifications.pdfA2- Trivalent-Tick-Fever-Vaccine-Specifications.pdf

	Combavac 3 in 1 Live Tick Fever Vaccine	*A. centrale , B. bigemina, B bovis*	U-F	http://www.daf.qld.gov.au/__data/assets/pdf_file/0003/53868/Tick-Feverhttp://www.daf.qld.gov.au/__data/assets/pdf_file/0003/53868/Tick-Fever-B- Combavac-3-in-1-Live-Tick-Fever-Vaccine-Specifications.pdfB-Combavac-3-in-1-Live-Tick-Fever-Vaccine-Specifications.pdf
South Africa	Frozen African Redwater Vaccine for Cattle (*B. bigemina*) Onderstepoort Biological Products Ltd. (OBP)	*B.bigemina*	U-F	
	Frozen Asiatic Redwater Vaccine for Cattle (*B. havis*)	*B. bovis*	U-F	
Uruguay	HEMOVAC C / Cibeles HEMOVACUNA/ DILAVE Miguel C Rubino	*A. centrale , B. bigemina, B bovis*	U-F R	http://www.cibeles.com.uy/es/?pg=vet_productos http://www.mgap.gub.uy/dgsg/dilave/Parasitolog%C3%ADa/Publicaciones/8_Epidemiolog%C3%ADa%20y%20perspectivas%20en%20el%20control%20de%20hemopar%C3%A1sitos.pdf
Brazil	EMBRAVAC®HEMOPAR/ Hemopar ERITROVAC®N2®/Hemopar ERITROVAC /Hemopar	*A. centrale , B. bigemina, B bovis*	U-F U-F R	http://www.catalogosnt.cnptia.embrapa.br/catalogo20/catalogue_of_products_and_services/arvore/CONT000fmt0bvzi02wyiv8003d0p31bvkqp1.html http://www.hemopar.com.br/index.php/produtos.html
Israel	Kimron Veterinary Institute	*A. centrale , B. bigemina, B bovis*	U-F	
Mexico	VACUNA CONTRA LA BABESIOSIS BOVINA/Cenid-Pavet- INIFAP	*B. bigemina, B bovis*	U-F	http://utep.inifap.gob.mx/tecnologias/1.%20Bovinos%20Leche/4.%20Sanidad/VACUNA%20CONTRA%20LA%20BABESIOSIS%20BOVINA.pdf

Commercial vaccines manufactured in Africa and Asia

In the countries targeted by the present monograph, only Ondesterpoort Biological Products (OBP) in South Africa produces vaccines for *B. bovis* (commercialised as Frozen Asiatic redwater) and *B. bigemina* (commercialised as Frozen African redwater) (**http://www.obpvaccines.co.za/products)**. They are both frozen vaccines and need to be kept on dry ice or in liquid nitrogen. Once thawed they will be effective for 30 minutes to 4 hours depending on the thawing method, and cannot be refrozen for a later use.

Commercial vaccines imported into Africa and Asia

To the best of our knowledge, none of the target countries, in the exception of South Africa, Zambia and to a limited extend Mozambique, practices vaccination.

None of the respondents from the countries of interest, replied that the babesiosis vaccine is being imported into their country.

Characteristics of Ideal Vaccine Candidates for Smallholders

Table 10: Target Product Profile (TPP) Bovine Babesiosis vaccine – Proposal:

	Attribute	Minimum (current available vaccine)	Ideal
1	Antigen	Immunogen with protective antigens of *B bigemina or B. bovis* that protects against respective infections	Immunogen capable of providing full protection in cattle against both *B. bovis and B. bigemina* infections
2	Indication for use	For active immunization of cattle & water buffaloes	For active immunization of cattle, water buffalos and all susceptible animals
3	Recommended species	Cattle, Water buffaloes	All *B. bovis and B. bigemina* susceptible animals
4	Recommended dose	2 ml	1 ml
5	Pharmaceutical form	Reconstituted injectable solution/suspension (freeze-dried vaccine) or ready to use solution (inactivated vaccine)	Ready to use solution/suspension
6	Route of administration	intramuscular	SC, Intramuscular or pour on
7	Regimen - primary vaccination	Single dose	Single lifetime dose

8	Regimen - booster	Single annual booster	Lifelong immunity after primary vaccination
9	Epidemiological relevance	Protection against all geographically distinct strains of *B. bovis* and *B. bigemina*	Protection against all geographically distinct strains of *B. bovis* and *B. bigemina*
10	Recommended age at first vaccination	Animals over 3 months: one injection	From 1-2 months of age
11	Onset of immunity	2-3 weeks following primary vaccination	One week following primary vaccination
12	Duration of immunity	At least 1 year	Lifelong immunity
13	Expected efficacy	To prevent disease & prevent mortality.	To prevent infection and transmission. No disease & no mortality in vaccinated animals after virulent challenge.
14	Expected safety	In animals under 6 months of age, a transient pyrexia reaction can occur. A transient nodular reaction of varying importance, may appear at the injection site, it progressively disappears within 1 to 2 months. Only vaccinate pregnant animals on emergency.	No post-vaccinal reactions at any age. Safe for pregnant animals. No carrier form in vaccinated animals
15	Withdrawal period	Nil	Nil
16	Special requirements for animals	Do not vaccinate un-healthy animals	Do not vaccinate un-healthy animals DIVA
17	Special requirements for persons	None	None
18	Package size	Multiple pack size from 5 doses	Multiple pack size from 5 doses

19	Price to end user	Not more than $0.50/dose	$0.20/dose at end user
20	Storage condition and shelf-life as packaged for sale	12 months at 4-8° C	24 months 4-8° C and/or 48 hours at 30° C
21	In-use stability	1 hour	24 hours

Overall conclusion for improved bovine babesiosis control through vaccination

In use over the last 30 to 40 years, live attenuated bovine babesiosis vaccines produced in splenectomised animals continue to be the only available form of vaccine to date. While they provide a good protection in immunized animals, there wide use is limited due to the complex and risky production process involving live animals, the requirement for cold chain and need for close veterinary or specialized supervision during administration. The poor results obtained with non-replicating vaccines and the discontinued efforts indicate that there may not be an alternative vaccine in the short to medium term.

None of the Asian countries have been producing or using babesia live attenuated vaccines.

Options that could be considered in the meantime are:

1. Further improvement of the current live attenuated vaccine: aspects to be considered would include cell culture, freeze-drying, combination of both *B.bovis* and *B.bigemina* in Africa.
2. Given the limited efficacy of inactivated vaccines, a vaccination program consisting of priming with inactivated vaccine and boosting with live one could be considered and evaluated as it may address the safety problems of the live vaccine, while aiding in the buildup of a solid long lasting immunity
3. There is a need to consider a broader use of the *R. microplus* tick vaccine in more regions in Africa and Asia; Although there is not yet evidence that the spread of the R. microplus tick in West Africa has corresponded to a worsening of Babesiosis, it will be critical to consider starting to deal with the problem before the parasite get established

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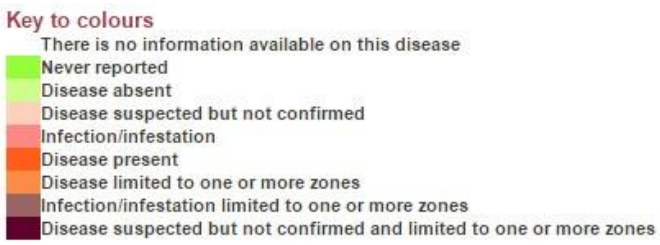
ANNEX 1: Additional data on disease presence and incidence

Reports to OIE on Babesiosis:

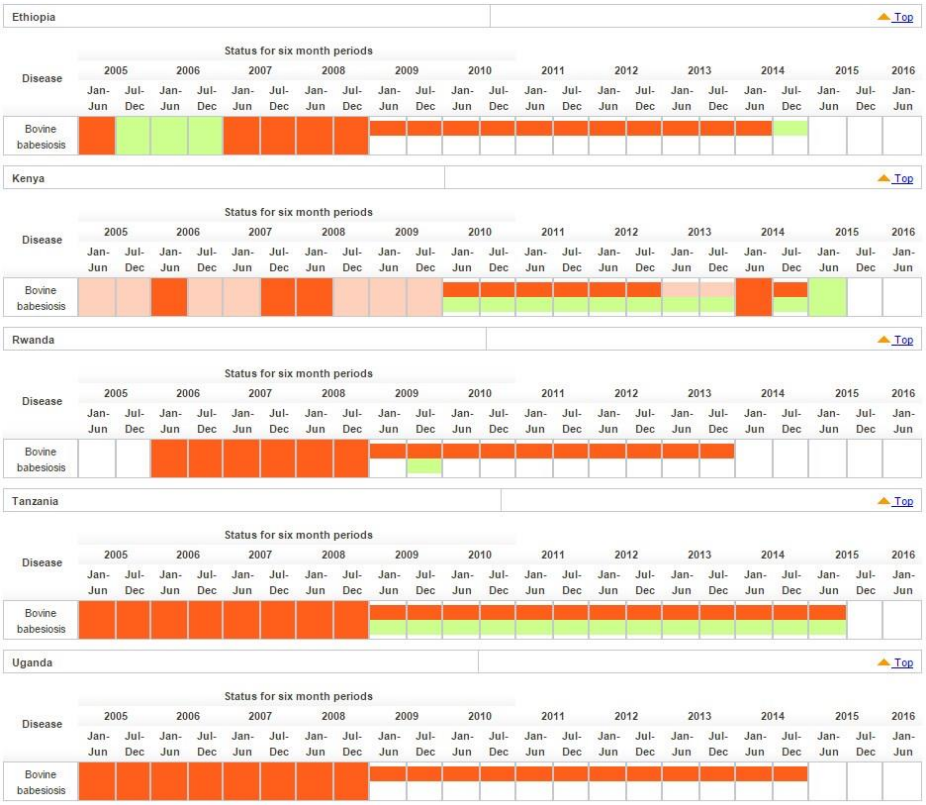


When different animal health statuses between domestic and wild animal population are provided, the box is split in two: the upper part for domestic animals, and the lower part for wild animals.

Babesiosis in Asia: Bangladesh, India, Indonesia, Myanmar, Nepal and Vietnam

Babesiosis in Western Africa: Burkina Faso, Ivory Coast, Mali and Senegal

Babesiosis in Eastern Africa: Ethiopia, Kenya, Rwanda, Tanzania and Uganda



Babesiosis in Southern Africa: Madagascar, Malawi, Mozambique, South Africa and Zambia